Wildlife Nutrition

A Publication of Canada's Accredited Zoos and Aquariums
Nutrition Advisory and Research Group (CAZA-NARG)

January, 2013

Happy New Year!

In this year's first newsletter, we are discussing plastic and food safety, fruit and amphibians.

The nutrition of amphibians may or may not be directly applicable to your job responsibilties. However, I think everyone will find that providing appropriate nutrition to captive amphibians is complex and challenging!

Deb McWilliams

CAZA-NARG
807-40 Vanier Drive
Guelph, ON, Canada N1G 2X7

519-823-4284
Info@caza-narg.ca
www.caza-narg.ca

In This Issue . . .

***"Wildlife Nutrition" is published three times yearly (January, May and September*)**

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Guelph, ON, N1G 2X7

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www.caza-narg.ca

The Use of Plastics and Food Safety

In my tours of zoo commissaries, I often see a wide variety of containers used for food storage and for feeding. Often, these containers can be inappropriate for food storage or for feeding and they are used only because of a lack of knowledge about the plastics coding system. Many times, zoo professionals will tell me they even use similar containers for their own food storage at home because of the belief that these plastics are safe to use.

Why are plastics a concern? All plastic is made from petroleum products. Chemicals that leach from products made of plastics are known to be carcinogens, endocrine (hormonal) disrupters, they can cause genetic damage in fetuses and, they can act as neurotoxins.

The "plastics coding system" was devised as a guide for the safe use and disposal of plastic containers. This system consists of the numbers 1 thru 7 inside a triangle made of arrows (below). Sometimes the arrows will have letters underneath the triangle, but usually the triangle is not labeled. Table 1 (page 4) explains what each number within the triangle can tell you about the plastic of the container and safe use of that container.

<u>Safe Guidelines for Use of Plastics:</u>

a. Heating of any plastic results in a break of the chemical bonds that hold the plastic together and chemicals will be leached from the container. Therefore, avoid heating food in plastic.
b. Do not use any plastic container, bag or food wrap for food or water that is discoloured, scratched or abraded. Damage to the plastic means the plastic has been eroded and chemicals will be leached into the foods. Scrubbing a plastic with an abrasive material will also erode the plastic. Materials that are discoloured, scratched or abraded also provide surfaces for the growth of bacteria.
c. "Microwave safe" does not mean chemicals do not leach from the container when microwaved (see a). "Microwave safe" means that the plastic will not melt (bubble) when used in a microwave and you are not at risk for a burn. All sources of heat will break the chemical bonds of plastics and result in the leaching of chemicals into food.
d. Avoid storing foods high in fat in plastic containers. There is a greater probabilty of chemical leaching from plastics with foods high in fat.
e. Plastic food wraps should not come into contact with warm or hot foods because of the potential for leaching into the food (see a).
f. Do not place hot foods into a plastic container, bag or food wrap. Chemicals are released from the plastic upon contact with hot food (see a). Heat breaks the chemical bonds of plastics and results in the leaching of chemicals into food. Let food cool before storing in any plastic material.
g. Do not wash plastics in a dishwasher because the heat of the dishwasher will break the chemical bonds that hold the plastic together and chemicals will be leached from the container (see a). Dishwashers are often used to sterilize plastic food containers because of the high temperature of the water. To sterilize plastics without using a dish washer, use a solution of 1 part bleach to 10 parts water and soak for a minimum of 5 minutes. Rinse well.

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. . . . . . Plastics Coding System (continued)

Number	Type of Plastic	Product	Safe Use of the Plastic
1	**Polyethylene Terephalate (PET**	Drink bottles, mouthwash, boil-in-bags, some cleaners, peanut butter jars	Safe for food, do not reuse. Cleaning causes leaching of chemicals such as di(2-ethylhexl) adipate (DEHA) known to be a carcinogen. Not safe in microwave.
2	**High Density Polyethylene (HDPE)**	Milk, detergent, nonfood oil, toys, trash bags, ice cube trays, food storage	Not safe in microwave. Do not reuse.
3	**Unplasticised or plasticized Polyvinyl Chloride (UPVC or PPVC)**	Food oils, food wraps, pet toys, teething rings, meat packaging, beverage pitchers, baby bottle nipples	***Not recommended for direct contact with food and avoid when possible.*** Cannot be re-used. Will produce toxins. Not safe in microwave. Recommended to avoid any #3 product.
4	**Low Density Polyethylene (LDPE)**	Squeezable food bottles, produce bags, bread bags, zip lock bags, baby bottle liners	Not safe in microwave
5	**Polypropylene (PP)**	Bottle tops, plastic bags, food wraps, yogurt and margarine containers, straws, spice containers	Not safe in microwave
6	**Polystyrene (PS) or Expandable Polystyrene (EPS)**	Plastic cutlery and foam packaging, egg cartons, meat trays, disposable cups and bowls, carry-out containers	***Not recommended for direct contact with food and avoid when possible.*** The plastic leaches into food. Do not use.
7	**Other, polycarbonate plastic, bisphenol A, nylon, acrylic**	Baby bottles, large water cooler bottles, meat trays, sport water bottles, plastic dishes	***Not recommended for direct contact with food and avoid when possible.*** The plastic leaches into food.

Dietary Challenge! Fruit: Wild versus Cultivated

The dietary challenge we will discuss this issue is the use of fruit in the diets of captive animals. I am frequently asked for recommendations on what fruit species to use in captive diets and how much fruit to feed captive animals. The awareness of, and diagnosis of, disease processes such as diabetes makes this a relevant dietary challenge when formulating diets for captive animals.

Before we begin, recall that carbohydrates are derived from plants and fruit is produced by plants – therefore fruit is composed of carbohydrates (among other nutrients). Recall also that the gastrointestinal tract (GIT) of carnivores are designed to digest proteins and fats – not carbohydrates. Therefore, the issue of feeding out fruit does not apply to carnivorous species.

In most zoological institutions in North America, the only fruit we can feed captive animals is "cultivated fruit". Cultivated, or domestic fruit, is any fruit that results from genetic manipulation and grown for palatability (taste) and appearance. The cultivated fruit eaten by humans is very different from the wild fruit eaten by animals. The table on page 6 compares some of the major nutritional differences between cultivated fruit and fruit grown in the wild

Wild fruit, as compared to cultivated fruit, provides a higher level of nutrition because it is high in fiber, protein and fat and low in sugar. As we can see in the table on page 6, if we feed cultivated fruit to a species that has evolved to survive on fruit grown in the wild, we have altered the nutrients provided to their physiology in major ways. In addition to a deficiency in nutrients, cultivated fruit that is high in sugar and provides minimal levels of protein, fat and fiber and can contribute to the development of disease processes such as GIT dysfunction and Type II diabetes. Page 6 has an illustration of this difference by comparing a wild banana variety and a cultivated variety of banana.

In general, the average protein level of wild fruits is 13.2% (Cook et al, 2000) and can be as high as 37.8 % (Glew et al, 1997) compared to < 5.5% for domestic fruits. Simple carbohydrates (sugars) average only 13.9% in wild fruits but are more than 34% in domestic fruits. Domestic fruit, especially when ripe, will have even higher levels of simple sugars than the wild fruits because the starch in unripe fruit converts to sugars during the ripening process.

In addition, wild fruit has a higher lipid content of an average of 4.9% compared to < 2.5% in domestic fruits. This fat is low in saturated fats.

Problems Wanted!!

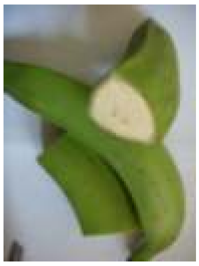
Each issue of "Wildlife Nutrition" will present and discuss a specific dietary challenge submitted by readers. Any aspect of the nutrition of captive wildlife will be considered for publication. The dietary challenge may be a question, situation or nutritional pathology. Questions re: body condition must be accompanied with a photo. The identity of the submitting individual and/or their organization will be confidential. Please submit to:

Wildlife Nutrition
info@caza-narg.ca

Dietary Challenge! Fruit:Wild versus Cultivated (*continued*)

So, what can we provide for captive species that normally would eat fruit in a wild diet? In general, we can use unripe cultivated fruit and vegetables. Unripe cultivated fruit will have higher levels of protein and complex carbohydrates (fiber) and lower levels of simple carbohydrates (sugars) when compared to ripe cultivated fruit. If possible, if an animal will eat unripe fruit, it is best to offer the unripe fruit.

Many cultivated vegetables can offer more nutrition with less simple carbohydrates (sugars) than cultivated fruit. Therefore, when possible, limit an animal to vegetables only. Vegetables, in addition to providing more nutrients, often have a sweet taste and will have a high rate of acceptability by most animals. For example, foods such as carrots, cooked legumes (beans), red peppers, yams (sweet potatoes) have very obvious sweet flavours. Other vegetables may have less of a sweet flavor, but still are widely accepted by many animal species because they provide a pleasant taste.



The above photos compare a wild, green banana (left) with a cultivated, green banana (right). Note the large seeds in the wild banana. The seeds in the wild banana increase the protein and fibre.

Cultivated Fruit	Wild Fruit
High to very high sugar content	Low sugar content
Low fiber	High Fiber
Low protein	High protein
Low fat	High fat
Small seeds or do not have seeds (this also decreases the fiber and protein levels of the fruit).	Seeds and often large seeds (increases dietary fiber and protein)

Amphibian Nutrition I: Introduction to Captive Nutrition of Anura and Caudata

Amphibian species in the Orders Anura and Caudata include frogs, bullfrogs and toads (Anura) and newts and salamanders (Caudata). Species in these orders have very simple gastrointestinal tracts (GITs), but providing appropriate nutrition to captives can be extremely complex because of ontogenetic dietary shifts and environmental toxicities. An ontogenetic dietary shift is defined as a change of nutrition niche according to life stage or developmental period. For example, a neonate might be carnivorous but gradually shifts (develops) to an omnivorous feeding strategy as an adult.

In addition to the complexities of ontogenetic dietary shifts and environmental toxicities, with few exceptions, dietary provision usually must include live prey under specific environmental conditions that include the appropriate temperature, humidity and light. Adding to the complexity of providing the appropriate nutrition to captive amphibian species is the lack of research on amphibian dietary needs.

The Amphibian Digestive System

An amphibian digestive system starts in the mouth. The tongue has taste buds capable of tasting bitter, salty, sour and sweet. The tongue of most anurans and terrestrial caudates is long, muscular and sticky ("sticky tongue pad") to capture prey such as adult insects, eggs (insect, fish and amphibian), insect larvae, small fish and, small rodents. Species such as Pipidae (clawed frogs), Sirenidae and, Proteidae (neotenic (juvenile) salamanders) that are fully aquatic do not have tongues and use negative pressure created by opening the mouth to pull prey in (buccal pump). Teeth are usually homodont (all the same type) and polyphyodont (continuously replaced) and, when present, are used to hold prey that is usually swallowed whole although forelimbs may be used to push food into their mouth.

The amphibian GIT is simple (short and without a cecum), digestion is enzymatic (e.g., pepsinogen) and, peristalsis (muscular action) and/or ciliary action moves food through the system. The stomach of amphibians is low in pH (acidic) and the enzyme pepsinogen is converted to pepsin to breakdown proteins into amino acids. Amino acids are digested by trypsin and carbohydrates are converted to simple sugars by amylase. Bile salts and further enzyme action break down fats in the small intestine where lipase converts fats to fatty acids and glycerides to glycerol. Amphibian livers change in size seasonally because it is the storage organ for glycogen and fat. The GIT ends in the colon and the cloaca (feces and urine). Nitrogen is excreted as ammonia by aquatic tadpoles and adult aquatic frogs excrete nitrogen in urine (urea). Most terrestrial frog species excrete nitrogen as uric acid.

Factors in Captive Amphibian Nutrition

Amphibians are ectotherms and several factors other than the digestive system must be considered for captive amphibian nutrition. These factors include the **integument (skin)**, **vision**, **olfaction**, **thermoregulation**, **metabolism** and, **immune functioning**.

The Integument

The diet of an amphibian affects the health of its skin. In turn, the functioning of its skin can also affect the health of the amphibian. The skin of amphibians has a multitude of functions important in obtaining and absorbing nutrients: it absorbs and secretes electrolytes and water; it has a respiratory function; it has a role in thermoregulation; it has a sensory function; and, it can be a source of nutrients.

Amphibian skin absorbs and secretes electrolytes and water. The permeability of amphibian skin allows cutaneous water exchange and it is essentially the mechanism by how they "drink". Specifically, the "pelvic patch" is highly vascularized and it is the site where most of the cutaneous water uptake occurs. The pelvic patch can efficiently absorb water from pools, droplets or soil. Terrestrial amphibians, however, are best adapted to obtain water from soil with lower water potential than can aquatic amphibian species. The exchange of water and electrolytes makes the integument also a part of the kidney system. Amphibian species in arid climates use lipids and waxes to coat their skin to reduce water loss.

Amphibian skin has a respiratory function. Respiration (breathing) in adult amphibians is by lungs and larvae respire via internal gills. Both, however, obtain and discharge significant amounts of oxygen and carbon dioxide via cutaneous (skin) gas exchange.

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Amphibian Nutrition I: Introduction (c*ontinued)*

Amphibian skin has a role in thermoregulation. The skin of amphibians can reflect or absorb heat. Adequate thermoregulation is important for GIT function and digestion.

Amphibian skin has a sensory function. Amphibian skin is very sensitive and can detect air currents, chemicals and electrodetection (electromagnetic detection) if the animal has a lateral line. These senses may be important in detecting prey and avoiding predators.

Amphibian skin can be a source of nutrients. Many amphibian species will eat their shed skin and/or feed young with shed skin.

Vision

The large eyes and binocular vision (both eyes used together) of anurans are essential for detecting prey (although amphibians also use olfaction when hunting). Tadpoles have smaller eyes, less binocular vision and rely more on olfaction for hunting. Anurans have colour vision equal to human ability.

Vision is also used to detect light and light of some level is needed to detect prey. For example, nocturnal amphibian species use low-intensity light for hunting.

Olfaction

Olfaction for prey detection is most important in tadpoles and in caudates who have smaller (or no) eyes and less binocular vision. However, all amphibian species have a Jacobson's organ (vomeronasal organ) and use it to differentiate hormonal states, individuals, eggs, breeding ponds and to detect prey and predators.

Thermoregulation

Amphibians are ectothermic and dependent upon the environmental temperature for physiological functioning including digestion and defecation. Extremes of temperatures (too high or too low) cause physiological stress and the animal will often stop eating and become inactive. Animals kept at temperatures too low for optimum physiological functioning may appear darker in color, they will be inactive and, they may not defecate.

All amphibian species, however, will attempt to moderate the environmental temperature in some manner if possible. The integument has mucous glands that cause evaporative cooling and can decrease body temperature. Using behavioural thermoregulation can also reduce or increase body temperature as needed by basking, seeking shade, using substrates, hibernating, estivation (torpor) or delaying activity until daylight or night fall. Therefore, all housing should provide a graduation of temperatures so the animal can use behavioural thermoregulation and move to hotter or cooler zones when needed. Aquatic species (larvae and adults) whose body temperature is dependent on the ambient water temperature depend upon behavioural thermoregulation.

The limited nature of captive environments does present a challenge to provide amphibian species with living areas that offer a selection of thermal areas. Pough (2007) states that individual temperature preferences and temperature tolerance of amphibian species can fluctuate daily, seasonally, by social interaction, with age and/or is dependent on the thermal history of an individual.

In general, when in doubt regarding the appropriate environmental temperature, recommendations are to start at a lower temperature within the species thermal neutral zone and gradually increase the ambient temperature. The thermal neutral zone (thermoneutral zone) is the temperature tolerance range of a species.

Metabolism

There is currently only minimal information available on amphibian metabolism. Both Donoghue (1998) and Huang et al. (2003) state that the equation for estimating daily kilocalorie provision for amphibians is $33(\text{weight in kg})^{.75}$. Donoghue, however, says the equation is estimating standard metabolic rate whereas Huang says the equation estimates metabolizable energy (ME). Therefore, the equation should be used as a starting point for daily kilocalorie provision then monitor for food intake, food refusals and body condition (e.g., weight loss or gain). In general, amphibians will eat more food if they are active and they will eat less or stop eating if inactive (assuming other factors such as illness, breeding, etc. are not present).

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Amphibian Nutrition I: Introduction (c*onclusion)*

Specifically for salamander species, those who are paedomorphic (have external gills for breathing in water and do not have metamorphosis) and metamorphic (use cutaneous gas exchange when in water and undergo metamorphosis) have metabolic rates that increase as the temperature increases. In water, paedomorphs and metamorphs have similar metabolic rates but metamorphs have a higher metabolic rate in water than on land. As well, male and female salamanders may have different preferences for temperature and basking schedules (factors directly related to metabolic function) and these preferences may also be related to phenotype (morphology).

Immune Functioning

Immune functioning can be a factor in amphibian nutrition because of the complexity of environmental factors in physiological functioning. The environment and husbandry will directly affect food intake, digestion, metabolism and production of endogenous hormones. A healthy frog is one who eagerly hunts prey and eats adequate amounts of food to maintain its physiology. A healthy amphibian will also have healthy skin that can function to protect against pathogens by producing peptide antibiotics in some species. Skin also contains pigments for camouflage and pheromone production for territorial issues and reproduction. This allows adequate, to optimal, communication with the environment and conspecifics to reduce stressors that can compromise immune functioning.

Immunologically-challenged animals may also have lost some aspects of fertility necessary for reproduction. For example, immune compromised northern cricket frogs (*Acris crepitans*) had reduced spermatic cyst diameter and germinal epithelium depth and these factors limited hatching rates (McCallum & Trauth 2007).

Amphibian Nutrition II: Adults

Adult amphibians are carnivores and hunt prey relative to their physical size. Smaller adult amphibians, for example, will hunt insects whereas larger amphibians will be able to hunt and eat fish, other amphibians, reptiles, birds, and small mammals.

Most captive feeding programs of amphibians do not feed amphibian species every day and prey is provided in limited quantities on feed days. This may or may not be appropriate given the sedentary lifestyles of most captive amphibians. However, given the high prevalence of nutritional pathology in captive amphibians and the feed intake rates of 100's to 1000's of a variety of prey daily in the wild, we may have to re-think our amphibian feeding programs and the environments we provide for them. This document cannot answer these – and other questions – but, the information presented in this document may assist change in a positive direction.

Adult Anura

The invertebrates commonly fed to captive amphibians are fruit flies (*Drosophila hydei* and *Drosophila melanogaster*), ants (various genera), crickets (*Gryllus* spp, *Acheta* spp), locusts (*Melanoptus* spp), springtails (*Collembola* spp), and blackflies (*Musca* spp). Beetle larvae such as superworms (*Zophobas* spp), mealworms (*Tenebrio* spp), and waxworms (*Galleria* spp or *Achroia* spp), brine shrimp (*Artemia* spp), water fleas (*Daphnia* spp), glass shrimp (*Palaemonectes* spp), various crayfish and, earthworms (*Lumbricus* spp), redworms (earthworm larvae), silkworms (*Bombyx* spp larvae), bloodworms (*Chironomidae* midge larvae), whiteworms (*Enchytraeus* spp), blackworms (*Lumbriculus* spp), and tubifex worms (*Tubifex* spp) are also fed. Vertebrate prey species are commonly used such as freshwater feeder fish (e.g., guppies, mollies, goldfish, smelt), and rats or mice (neonates to adults).

Captive feeding often does not develop beyond insectivory using a few species, even for large amphibian species. However, it is recommended that feeding a variety of prey not only increases the activity and welfare of the captives, it will provide optimum nutrition. In addition, prey must be raised and/or kept in nutrient-rich environments to provide the amphibian predator with optimum nutrition.

Adult Caudata

Aquatic salamanders in the wild (larval salamanders, juveniles and adult salamanders) prey on leeches, snails, crustaceans, insect larvae, small fish and other amphibians. Many of these species are cannibalistic. Brine shrimp (frozen and dried), whole minnows and fish fillet pieces are often fed in captivity. Filets of fish are sometimes used, but we must consider filets to be nutritionally deficient because they are only muscle meat and lack many of the nutrients available in whole carcasses.

Terrestrial salamanders in the wild prey on invertebrates (earthworms, slugs, insect larvae and nymphs, adult insects, arthropods). In captivity, earthworms and beetle larvae (*Tenebrio molitor*) are fed, especially to Ambystoma and Plethodon species.

Dietary Protein and Fat

As carnivores (meat-eaters or insectivores), amphibian diets will naturally be 30% to 60% protein (metabolizable energy – ME). Or, in other words, protein is 30% to 60% of the calories taken daily. For these species, dietary protein is used as an energy source. Carnivores cannot digest and use carbohydrates for energy.

As carnivores, the dietary fat fraction of amphibian diets will naturally range from 40% to 70% of calories. Dietary fat is also used for energy.

Carbohydrates

With a diet consisting of 30% to 60% protein and 40% to 70% fat, there is only a negligible amount of carbohydrates (plant material) in a carnivore's diet. Carnivores fed diets with an excess of carbohydrates develop several nutritional pathologies including intestinal blockage.

Vitamins and Minerals

The available information on the dietary vitamin and mineral needs of amphibians is limited to calcium metabolism. While we do not know the vitamin and mineral needs of amphibians, nutritional pathologies are prevalent in captive amphibians and many of these pathologies are linked to inappropriate vitamin and mineral supplementation.

As mammals, we may tend to focus on the oral ingestion of nutrients. With amphibians, however, some nutrients are obtained from the environment passively via exchanges between the integument and the environment. Calcium, for example, is one mineral that can be absorbed via the integument in adult amphibians (Kingsbury and Fenwick, 1989) and via the gill surface in anuran tadpoles (Baldwin and Bentley, 1980).

As always, meeting the vitamin and mineral needs of an animal should be done in view of the environment where the animal's physiology has evolved. For example, neotropical amphibians may have evolved in environments with comparatively low levels of minerals. This may indicate that their physiologies are efficient at obtaining, using and storing micronutrients. Such efficiency may predispose these species to o developing vitamin and mineral toxicities.

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Amphibian Nutrition II (*continued*)

Feeding Schedule

The timing of feeding appears critical for most captive amphibians. Diurnal species should be fed in the morning and nocturnal species should be fed at dusk. For some terrestrial species, misting immediately before feeding may stimulate feeding because some species (e.g., Dendrobatidae) are more active after a rainfall. Food that is not eaten within a reasonable time (usually 1-2 hours if fed appropriately) should be removed to prevent spoiling, loss of nutrients, and potential trauma to the amphibian (e.g., attack by prey).

Water

Maintaining an osmotic balance with the environment is necessary for survival. Amphibians urinate or lose moisture via the integument at about 33% of their body weight daily, therefore are at risk for dehydration. The skin of many amphibian species can absorb water and cells of the integument can also actively pump salt for maintaining an osmotic balance.

Whether an amphibian is an aquatic or terrestrial species, hydration is important to maintain health and physiological functioning. The use of municipal and well water for captive amphibians requires some water management to assure the well-being of the animals. Recommendations include:

1. Let water stand for 24 to 48 hours before use to allow chlorine to dissipate.
2. Cleaners containing ammonia, chlorine and phosphates should not be used in the exhibit and/or on items to be used in the exhibit.
3. Limit nitrite levels to <0.1 parts per million (ppm). (Whitaker, 2001; Banks et al., 2008)
4. Limit nitrate levels to (<1.5 mg/L (Banks et al., 2008).
5. Flush the water system to remove standing water that may have leached chlorinated biphenyls or metal into the water.
6. Metals known to be toxic to amphibians include aluminum, antimony, arsenic, cadmium, copper, lead, manganese, mercury, molybdenum, silver and zinc (Blaustein et al., 2003; Browne et al., 2007).
7. Carbon dioxide levels should be < 6 mg/L.
8. Uneaten food should be removed from the water immediately after feeding is completed by aquatic species.
9. Unionized ammonia levels should be < 0.02 ppm (Diana et al., 2001).
10. Latex gloves can be lethal to tadpoles (Sobotka & Rahwan, 1994).

A potential problem during winter with municipal and well water is a build-up of gases (gas supersaturation) that can cause "gas-bubble disease" (the bends) in aquatic amphibians because cold water can hold higher levels of dissolved gas. The build-up of gases can include nitrogen, carbon dioxide and/or oxygen. The problem is easily solved by aeration allowing the gases to dissipate and/or warming the water to decrease its ability to hold high levels of gas.

Cannibalism

Cannibalism does occur among many amphibian species. In general, housing by size (e.g., small with the small) at all life stages will reduce or eliminate most cannibalism. However, research indicates that some cannibalism may be necessary for development and survival.

Nutrition and Reproduction

In general, about two months prior to breeding season, captive amphibians should be fed ad lib with high quality feed (Browne and Zippel 2007). Consuming large amounts of prey are common in the wild prior to breeding season and during seasonal increases of invertebrate populations. However, the provision of food should return to normal levels after breeding to prevent obesity.